ARS SPECIAL REPORT

Plant Light-Growth Discoveries

From Photoperiodism to Phytochrome

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Growth Through Agricultural Progress

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FOREWORD

Twentieth century man is managing plant growth with success undreamed of by farmers and gardeners before him. But scientists who explore plant life are aware that far greater success is ahead--when they can explain precisely how and why plants grow as they do on a sunlit earth.

Major progress toward this goal comes through discoveries disclosing the sun-operated clockwork in plants that controls each change in the life cycle from seed germination to fruiting.

Agricultural leaders will increasingly need information on this light-growth research and its implications, as background for appraising and explaining many of the new developments in plant science.

Information in this report was provided by the Crops Research Division and the Soil and Water Conservation Research Division of the Agricultural Research Service.

PLANT LIGHT-GROWTH DISCOVERIES...
From Photoperiodism To Phytochrome

Getting out of living plants a chemical that triggers their growth changes, from seed to fruiting, has brought us closer to a new era in agriculture--a time when nature's mechanisms will be understood well enough to avoid many of man's uncertainties over what to expect of a planted crop.

When the U. S. Department of Agriculture announced this breakthrough achievement in September 1959, the news introduced a new name: phytochrome, from Greek words meaning plant color. Scientists in the USDA Agricultural Research Service coined the name to fit a chemical they had detected in plants in the early 1950's, and can now separate from plants and examine in almost pure form.

The chemical, a light-sensitive blue pigment, is so tiny in amount that it gives no tinge of blue to a growing plant's color. Yet reaction of the blue substance with the sun is potent and direct. Light and darkness have other roles in the life processes of plants. But phytochrome's go-stop signals direct a plant's progress from one stage to the next as if a master valve were operating.

Revelations of phytochrome's existence and workings give new meaning to discoveries that began in 1918, when two USDA scientists learned that some plants wait for short days to form seed. They also found that plants differ in the day lengths that lead to seed formation, flowering, and other transitions in a life cycle. The discoverers coined a name for their new-found principle of growth: photoperiodism, to mean "the response of organisms to the relative length of day and night."

Light-growth discoveries have far-reaching importance for farmers, florists, gardeners, and, in fact, plant growers everywhere. A science-minded public may be expected to take increasing interest in this space relationship of the sun and earth's plants.

HOW LIGHT-GROWTH KNOWLEDGE SERVES AGRICULTURE

Since all plant growers deal with some specific plant in its environment, fragments of photoperiodic knowledge have been put to agricultural uses almost from the time the initial discovery was reported in 1920.

Florists First in Commercial Crop Application

Florists were the first commercial growers to take interest in technical aspects of photoperiodic science to improve their crop management for timed markets. For nearly 40 years, they have controlled day-night length with artificial light and shades of opaque cloth.

They know that expected blooming of some flower crops can be thrown off if night is not a complete blackout. Even a street lamp near a greenhouse has been known to delay a poinsettia crop from blooming

in time for Christmas. Home-grown poinsettias may similarly fail to keep a Christmas blooming date if some house lamp cuts their dark hours too short.

Florists in the South have extended artificial lighting to outdoor acreages of autumn and winter chrysanthemums, where a cash return per square foot would justify the cost--to delay flowering for timed markets.

Field Crop Growers

Outdoor light control may always be too expensive for much use on field crops. But farmers can make good use of photoperiodic knowledge in other ways--to outwit some adverse growing conditions, for example.

When rains prevent planting on schedule, some crops can be planted several weeks late and still catch up well enough to mature a crop. Soybean progress is so strictly controlled by day-length changes that a succession of plantings even of the same variety tends to mature only a few days apart.

Photoperiodic knowledge was a national asset in World War II when rainy spring weather in 2 years threatened much-needed soybean production. Farmers who thought it hopeless to try for a belated crop were reassured by plant scientists that they had time to get a worthwhile yield. Production per plant might be smaller than if sowing had been at the best time.

Knowing that some crops shift growth stages in close step with the sun gives farmers a wider choice of planting dates in favorable weather, as well as emergencies, to gain some advantage. They can time the planting of certain crops to minimize weed competition, or to allow for more vegetative growth by a forage plant.

Some of the newer knowledge of what light does in launching seed germination may help farmers eventually to fight weeds in a new way. Such use of light depends on technical findings explaining complex relationships of light, temperature, moisture, and other environmental conditions. But with enough knowledge, it may become practical to induce weed seeds to sprout all at once underground, and then destroy them quickly with chemicals.

Experimental Growers and Nurserymen

Plant breeders and other experimenters control light as the florist does, indoors and out, and in more varied ways.

Day-length management enables breeders to bring different varieties--with different photoperiod requirements--to bloom simultaneously for crossing. It enables them also to grow more generations of seedlings a year, thus providing new varieties faster for farmers' needs. These techniques have served plant breeders since 1922, when they found that in one season they could grow two crops of some wheat varieties in a greenhouse and still have time to add a third crop in the field.

When breeders are choosing among unfamiliar varieties as prospects for crossing, they often grow seedlings under different day-length conditions to judge suitability for different agricultural regions. One main reason for this country's success in developing and making use of improved crop varieties is that breeders have paid close attention to the reactions of candidate varieties to day length. Proportions of light and darkness in each 24-hour cycle are one of the most predictable features of a plant's environment, because they lengthen and shorten in the same dependable pattern year after year. Favorable proportions are so important to some crops that a variety well suited to one farming locality may not be nearly so productive in a latitude as near as 100 miles to the north or south.

Almost any nurseryman who propagates woody or other plants can make good use of photoperiodic principles. And the same is true for any crop grower who tries crossing or other experimental work.

The woody plants that grow slowly have shown particularly striking responses to light control in recent photoperiodic research. And young trees and shrubs are readily manageable.

Basic Advantages the Most Important

Beyond recognized uses--and more could be cited--the technical findings on light-growth relationships are important first and foremost for basic knowledge of how and why a plant develops as it does in a given environment. Only when nature's laws and intricate processes are well understood can man hope to adjust agricultural practices to plants and to modify plants to his advantage in a truly scientific way.

THE USDA ROLE IN PHOTOPERIODIC RESEARCH

The U. S. Department of Agriculture has worked at photoperiodic research continuously and with an increasing sense of purpose from the discovery of the concept. The Agricultural Research Service's Plant Industry Station at Beltsville, Md., has become a world center for pioneering advances in understanding this phase of plant growth.

The initial discovery came from a side-line interest of two gifted plant specialists, W. W. Garner and H. A. Allard, whose main task was to deal with tobacco crop problems.

Although they recognized the importance of their discovery at once, they waited 2 years and learned much more about day length and plant growth before they published their first announcement: A 53-page account of the photoperiodic concept and their experiments in a scientific journal. Scientists around the world showed keen interest. Garner and Allard continued this line of study in the Department for about 20 years.

When the Bankhead-Jones Act of 1935 provided funds for extending basic agricultural research, one prompt action of USDA was to set up a research unit to work entirely on plant growth and light relationships.

Today, the headquarters for this research at Beltsville are in one of the pioneering research laboratories that the Department established in 1957.

USDA's experience in photoperiodic research in 40 years has attracted a considerable number of staff scientists to join teams making light-growth experiments. Three, in particular, have been leaders since the Garner and Allard era:

Dr. M. W. Parker, plant physiologist, worked on many light experiments from 1936 to 1952, and is now director of the Crops Research Division in the Agricultural Research Service. Dr. H. A. Borthwick, another plant physiologist, joined in the photoperiodic research in 1936. He now leads the group specializing in this work in a pioneering laboratory designated for the study of "light as an environmental influence on plant life." Dr. Sterling B. Hendricks came to the USDA as a soil chemist, and by 1944 began working with Borthwick. Hendricks is currently leader of a pioneering group studying the mineral nutrition of plants, but he continues to work actively also with the photoperiodic team. He has been the key man in discovering and separating from plants the vital phytochrome that triggers growth changes.

THE DISCOVERY ROAD

The straightest short-cut to catching up with photoperiodic discoveries is to start at 1918 and think forward with scientists through major advances.

A few selected experimental projects by ARS plant pioneers working as teams, and in some cases with cooperating agencies, can suffice to show how clues and their meaning have led forward from one landmark to the next on the discovery road.

Four Milestones

Major milestones thus far are these:

1. The take-off point, that photoperiodism is a natural law of growth.
2. Finding that plants use one part of sunlight, red, in launching growth changes.
3. Detecting a hidden substance that works reversibly, operating go-stop signals under red and far red.
4. Bringing from hiding this chemical substance, and proving that it is a plant's means of sensing time and responding to season.

HOW PHOTOPERIODISM WAS DISCOVERED

Three Tobacco Plants and a Few Soybeans

Like some other laws of nature, photoperiodism was discovered in simple circumstances. On July 10, 1918, while armies in France were readying for the Second Battle of the Marne, two scientists in Virginia at 4 o'clock in the afternoon took three tobacco plants and a box of soybean seedlings into a small, crude shack. They had placed the shack in a field at the USDA experimental farm, which was then across the Potomac from Washington, D. C. They needed a dark room to try the latest of their uncounted attacks on one of nature's obstinately held secrets. This day they were making scientific history.

Garner and Allard as a crop research team had puzzled for years over the question: Why did Mammoth tobacco grown in Maryland continue vegetative growth so long in summer that it bore giant leaves and bloomed too late to form seed before frost? Maryland growers who planted Mammoth variety had to grow seed in a winter greenhouse. Biloxi soybeans similarly failed to form seed in this area.

Testing probable causes for this seeding habit, the two scientists had eliminated temperature, moisture, fertilization, and light intensity. Now, they were trying day length. They would give test plants 17 hours of darkness--from 4 P.M. to 9 A. M.--and then put them outdoors for a 7-hour day in July, like the days in late autumn and winter when Mammoth tobacco bore greenhouse seed crops. After a few days, the test plants bloomed and then formed seed.

Armed with a new concept--that some plants are short-day plants--the two scientists tested more plants, including cabbage, carrots, lettuce, ragweed, and wild violets. They found that plants differ in the proportions of day and night that lead to seed formation, and that these proportions also govern flowering, stem lengthening, and other growth transitions.

Accounting for Earth's Plant Distribution

The initial report by Garner and Allard in 1920 marshaled evidence that temperature, water, and light intensity could no longer end the list of main external influences governing where and how widely the earth's plants can grow. Scientists were asked to consider a new underlying cause--the relative length of days during a growing period.

The report indicated a 4-way grouping of plants that explained for the first time why vegetation differs with latitudes and seasons:

- Plants that normally flower in late spring or in summer respond to long days and may be called <u>long-day</u> plants.

- Conversely, plants that normally flower in the autumn or winter respond to short days and may be called <u>short-day</u> plants.

- A third and large group can flower and fruit under a wide range of day lengths. Garner and Allard later called these <u>indeterminate</u> plants.

- Plants tested and assigned to each group had varying day length requirements. Some were so near to a 12-12 hour response that they might be separated into a fourth group and were later given the name <u>intermediate</u>.

Some close plant relatives, such as varieties in the same species, fitted into different photoperiodic groups.

Photoperiodism as a Life Principle

The Garner and Allard report introduced a life principle so broad that it operates extensively over our sunlighted world; so basic that without it plant and animal life as we know it could not exist; and so complex that the related parts of the operating mechanism are just beginning to be fitted into place.

Some animal life changes are under photoperiodic control, as Garner and Allard indicated. Photoreactions have helped to explain some of the reproduction, dormancy, and migration habits in animal life.

Plant forms from the highest down to the tiny algae are believed to time growth changes by day length. But the system has been studied first and foremost in seed plants because of their major importance to agriculture.

Although an indeterminate plant, such as the tomato, can fruit from the tropics to the Arctic, such plants contain the same light-responding mechanism as short-day and long-day plants, though operating less dominantly. The tomato at maturity can develop skin color only in close compliance with photoperiodic behavior.

CLARIFYING THE ROLES OF NIGHT AND LIGHT

Giving Darkness its Due

For nearly 20 years after photoperiodism was discovered, plant scientists assumed that plants timed growth changes by some means of measuring daylight. But experiments devised in 1937 showed that plants measure the dark period in a 24-hour cycle, not the light. That is, a plant starts a new growth stage on a signal developed by clocking hours of darkness.

Remarkable features of this timing of darkness were demonstrated by soybean seedlings, which served as convenient and highly responsive test plants.

The soybean's dark-timer proved itself a precision instrument that could recognize a few seconds of light as an interference. A minute of artificial light at midnight each night was enough to keep young soybean plants from flowering. The dark-timer inside the plant disregarded the insufficient dark period prior to the light interruption and started clocking all over the instant darkness resumed.

When a light interruption was shifted toward the beginning or end of the dark period, the test plants responded less consistently or even ignored the light break entirely. Their maximum response came about midway in a long dark period. The exact reason for this difference has not yet been explained.

The dark-timer was sensitive to interruption from even faint light. This accounts for some bloom failures such as those cited on page 1.

In one test, soybeans were induced to bloom by keeping a single leaf on a plant in total darkness for 16-hour nights, while all the rest of the plant had inadequate darkness of 8-hour nights. Such a spread effect from a small control center indicated an intricate channeling inside the growing plant.

Tests of many plants besides soybeans confirmed the clocking of darkness as a prevailing plant characteristic. A long-day plant like barley has been brought to flower in winter greenhouse conditions by a little midnight lighting to keep its dark periods short. Barley's response to interrupted darkness is not so fast as the soybean's, but as little as an hour of light separating parts of a dark period is enough to promote flowering in most barley varieties.

Light Periods Not Clocked

To learn whether plants also could clock their light periods, the scientists reversed the midnight treatment. But no plant altered its flowering time when a day length was interrupted with brief darkness, even midway at noon.

A Boon to Growers

Before breaks in dark periods were tested, plant growers had taken care to extend day length by using electric current for some hours either before sunrise or after sunset, to hold back flowering in short-day plants, sometimes continuing this delaying treatment for many weeks. Chrysanthemums were almost the only commercial crop that justified the extra expense in light bills. Growers of chrysanthemums and some other greenhouse crops welcomed the evidence that midnight light would give more dependable results and at a bargain rate of an hour or less of current per night.

A Note on Terminology

Since plants clock darkness, not light, it might seem realistic at this point to adopt the adjectives "short night" and "long night" to describe plants from the angle of their photoperiod timing.

However, the Garner and Allard discoveries are so much a part of all follow-up findings, and so often referred to, that basic terms they introduced continue to be used for consistency. Plants, therefore, are still commonly called long-day and short-day types.

More Importance for Light

Giving darkness its due has not relegated light to any minor role in growth changes. During light periods, both the intensity and the color of light are important to photoperiodic responses.

High intensity of light during all or part of the light periods promotes starch formation. Experiments have shown that during darkness a plant's photoperiodic mechanism draws on the carbohydrate reserves, and in fact, the mechanism cannot operate if these reserves fall too low. Furthermore, in most plants, bright light speeds the initiation of a growth change. Soybean plants have started to flower after only three or four cycles in which 14-hour nights provided adequate darkness and each 10-hour day provided bright light.

Plants differ in their favorable responses to light intensities, as to other factors that influence their photoperiodic reactions. Knowledge of these light intensity effects is valuable for predicting how well a variety may be expected to yield in a given latitude and climate. Adequate intensity of light is rarely lacking for plants that flower and fruit in days of long, bright light. Daylight intensity is sometimes less dependable for plants that mature in short days. Indoors, light intensity can be manipulated for many significant effects.

The color of light is so important to photoperiodic plant responses that discoveries about this have been major advances in understanding photoperiodism--described in following sections.

SINGLING OUT RED

Since 1945, scientists have been able to concentrate on red as the color of light--the portion of white light--vital to plant growth changes.

The importance of red was established by treating plants with waves of different lengths that make up each color band in the spectrum, from violet at about 4000 A. to the visible limit of red about 7200 A.[1]

An Unusual Light-Breaking Instrument

Treating plants with light of individual rainbow colors was achieved when ARS scientists built an unusual spectrograph at Beltsville in 1944. Since materials were short in World War II, the instrument was made mainly of scrap. Yet its size and effectiveness have rarely been duplicated, and it continues in use today.

Main features combined in the instrument were a carbon arc lamp producing light strong enough to throw a beam far down a windowless room, and two large glass prisms that dispersed the white light into broad bands of separate colors on a screen.

The experimental advantages gained were these:

- The rainbow-colored band was so long that a row of 14 small pot plants could be irradiated at once, to induce their growth responses to separate colors of light. The entire color band was over 5 feet long and 3 inches high.
- The focal length of the instrument--nearly 33 feet--gave remarkably pure light of the various colors of the spectrum. There was so little scatter effect of one color band infringing on another that radiation impurity was a mere five-hundredths of one percent.

Light of Many Colors on Plants

For systematic spectrum testing, two short-day plants were chosen--the soybean and the cocklebur--because both had been used extensively in photoperiodic research.

To range a maximum number of pots on a bench within the spectrum rays, all foliage of each seedling was removed except a single leaf. In each 24-hour cycle, all plants received a dark period totaling enough hours to start flowering--but with a brief interruption around midnight, when each plant received light of a designated color. Figure 1 shows plants ranged for such a test. The single leaf of each plant was fastened to the screen, so that the exact wave lengths of light received could be recorded. Throughout 2 hours around midnight, when a light interruption

[1] A. = Angstrom unit. One such wave length unit is a hundred millionth of a centimeter.

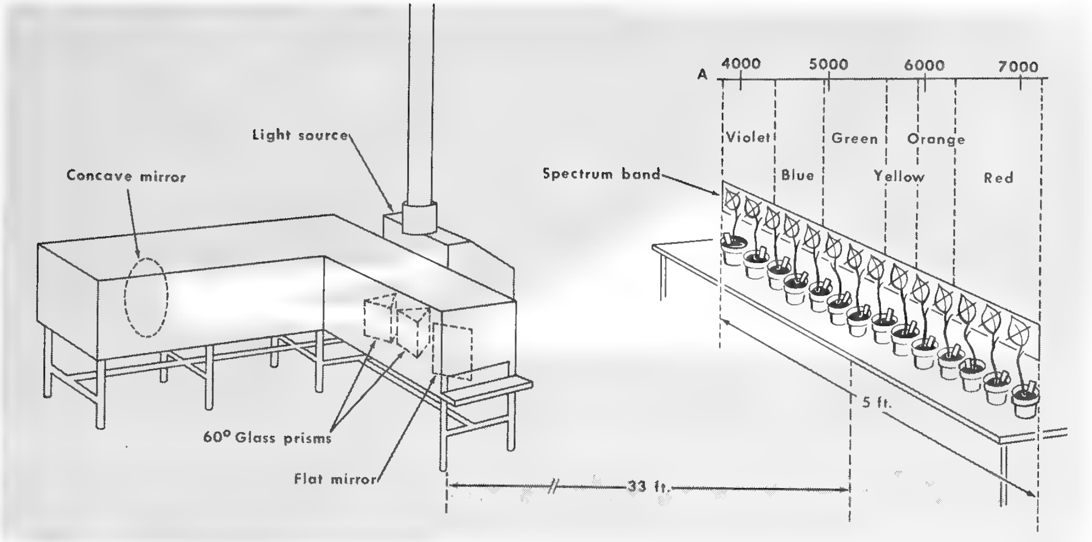

Figure 1.--In this diagrammatic drawing, room length has been much foreshortened to permit showing some details of the large ARS spectrograph and the relationship of the instrument to plants irradiated by it. In the initial experiments that showed the importance of red, each soybean test plant when ready to flower had all foliage removed except one leaf. This leaf was fastened to a screen for brief irradiation during each night photoperiod--to record flowering response to separate colors of spectrum light.

would have maximum effect, the scientists each night set up group after group of plants to be irradiated for different test times from seconds to a half-hour.

A red light interruption of even 30 seconds caused soybean and cocklebur plants to start clocking all over when dark was restored. This was evident because these plants never registered enough darkness to signal flowering time when the test conditions were continued for many weeks.

Plants receiving an interruption of other colors of light either flowered as if no light had appeared, or were affected at most only slightly.

When the outstanding effectiveness of red was confirmed with varied kinds of plants, the region of the spectrum ruling photoperiodism was mapped in the wave area from about 5800 A. to about 7200 A.

Clue to an Unknown Chemical

These spectrum tests gave scientists the first strong clue to the plant's responding mechanism. They had learned that both long-day and short-day plants respond to the same wave lengths of light. This could only mean that all of these plants, so different in photoperiod requirements, must contain the same responding mechanism. There must be in plants some unknown chemical substance strongly influenced by the red in sunlight.

REVERSIBILITY OF THE MECHANISM SHOWN BY RED AND FAR RED

Concentrating on the red in sunlight, photoperiodic pioneers were led to their next major milestone--finding that the unknown chemical works a reversible shift.

Seeds were the part of the plant to disclose how the hidden chemical develops signals. Seed research provided photoperiodic specialists with the leading clue of an experiment in which seed refused to germinate under light rays of 7000 to 8000 A. This suggested that light in the far red part of the red band gave "wait" signals resembling darkness, worth special investigation. In the early 1950's, the photoperiodic research team was joined by Dr. Eben H. Toole, seed specialist, to begin systematic studies of red, far red, and darkness.

The reversible action of the plant's growth-triggering chemical was demonstrated strikingly by responses such as those of small lots of lettuce seed. First, 30 seconds of red light and then darkness caused a few of the lettuce seed spread on a wet blotter to sprout in 2 days. Prolonging the red light exposure up to 16 minutes induced general sprouting in a lettuce seed lot. Next similar lettuce seeds that had been given enough red to assure readiness to sprout were given brief far red for intervals up to 16 minutes--and this brief far red halted sprouting up to almost 100 percent. Then, even more striking evidence: The seed response was pushed back and forth by alternating red and far-red light, and the last light always ruled, as shown in figure 2.

Such rapid sequences of light are highly artificial, not duplicated in nature. The importance lay in demonstrating the existence of a reversible mechanism hidden in a plant. Putting it through paces showed what it could do. The paces showed that far-red can do what darkness does, and sometimes more. Far red gave signals so potent that they inhibited

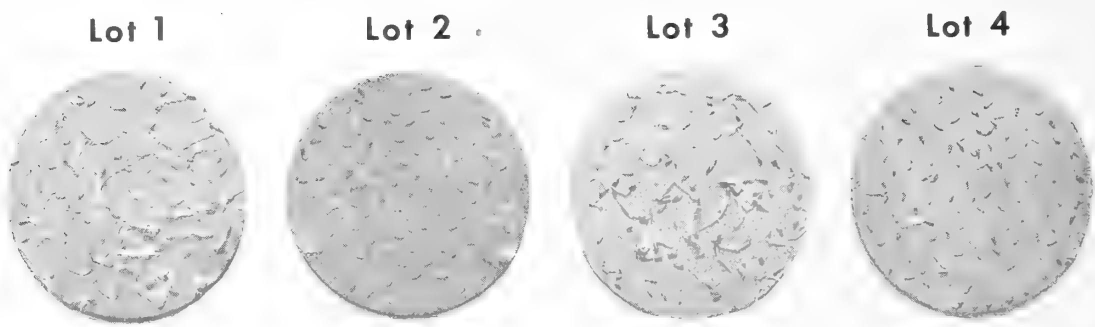

Figure 2.--These lettuce seed provided the dramatic and conclusive evidence that the growth-triggering chemical in plants reacts reversibly to red and far-red light. In preceding tests, the scientists induced small lots of lettuce seed to sprout by applying red light briefly, and held back sprouting in similar lots by applying far red. The four lots pictured show the striking final test results of applying red and far red alternately: In lot 1, red induced sprouting. In lot 2, red followed by far red prevented sprouting. In lot 3, a red, far red, red treatment induced sprouting. In lot 4, a red, far red, red, far red sequence prevented sprouting. The light-sensitive chemical reacted rapidly to each form of light--and the last light always ruled. In these laboratory tests, all seed lots spread on wet blotters had the same preparatory management of dark storage at 20° C. (68° F.).

germination in some seed that normally germinate in darkness. The tests were extended to many kinds of plants in growth stages, and showed that the same reversible action controls stem-lengthening, flowering, and other changes in plant development.

This evidence necessitated extending the band of the spectrum that governs photoperiodic responses. Photoperiodic researchers assign the name "red" to light wave lengths from about 5800 to 7200 A., and "far red" to wave lengths from about 7200 to 8000 A. Far red is a borderland including the longest waves of faintly visible light and extending beyond to include some infra-red waves that man's eye cannot see.

PREDICTING THE NATURE OF THE GROWTH-TRIGGERING CHEMICAL

At this point, the hidden light-sensitive chemical's characteristics and action could be predicted to a remarkable extent. The main clues and deductions were these:

The chemical is in all kinds of plants, working reversible shifts to control orderly growth changes.

The chemical is a pigment because it reacts to light in color fashion. It is blue, or just possibly green-blue, because only such a pigment would absorb as it does the complementary red and far-red waves of white light. It would not be closely related to chlorophyll because green chlorophyll reacts strongly to blue as well as to red.

The triggering pigment is in extremely tiny amounts, because albino plants, that have a bleached look for want of normal color, respond reversibly to red and far red, showing presence of the blue pigment.

The chemical is a dual form--a type that occurs widely in nature. In such chemicals, each molecule contains some atoms arranged in reverse pattern, like a right hand and its mirror image or a left hand. Red light absorbed by the molecule would move these atoms in the blue pigment into the right-hand or active form that triggers a growth change. Far red, and also darkness, would push the pattern into the left-hand form that inhibits a change. In sunlight both red and far red would be constantly pushing the atoms opposite ways, but with red more commonly prevailing.

The concentration of the pigment's active form has to reach a definite level before a growth change can start. Awaiting the right concentration, the plant would keep to its current activity, such as bigger leaf production in the vegetative stage.

BRINGING PHYTOCHROME FROM HIDING

The outstanding advantage of getting the growth-triggering pigment into the open was achieved in 1959. One of the research group named it phytochrome. Within a few months the group was able to announce how this basic part of a plant was located and separated, and to describe new information gained by working with it directly.

Especially impressive is the fact that the chemical in a test tube has not upset any of the deductions about it, formed from observing the reactions of growing plants and their seed.

A Pigment Detector's Report: Phytochrome Here

A sensitive instrument for measuring small amounts of pigment made it possible to locate phytochrome's presence and to report whether it was in active or inhibiting form. This single-beam spectrophotometer was devised by Karl Norris, research engineer of the USDA Agricultural Marketing Service, for investigations of plant color and market quality. The instrument scans a selected region of the spectrum, casting light of changing wave length on a plant or tissue. At each wave length, it measures the light transmitted through the dense material, registering the absorption by ink markings on a chart.

Using this instrument, the research team applied successive red and far red to 6-day-old corn seedlings and got direct evidence of phytochrome's presence and reverses. Seedlings for these tests were grown in the dark to prevent their developing green chlorophyll, which reacts to red and might interfere with the evidence. The young plants own seed gave them food for 6 days of growth with no light.

Ground-up tissue from corn plants and liquid extract from the tissue were tested also. And to locate phytochrome in varied kinds of plants and at different stages, the instrument was directed on test materials of 20 plant species. These included the stems and flowers of other grasses besides corn, spinach leaves, cauliflower's white florets, and fruits of avocado and zucchini squash.

A Rapid Detector's Assay Report

A second instrument built by AMS engineers made it possible to assay more exactly and quickly the tiny amounts of phytochrome distributed in plant tissues. This dual-monochromator spectrophotometer has much more sensitivity than the single-beam instrument and it gives wanted information instantly, without the mathematical computation necessary to relate the data on wave length effects when the single-beam instrument is used. In figure 3 a bean seedling is in position for a phytochrome assay.

Assays with this instrument indicate that phytochrome amounts at the most to a millionth of a plant's total weight. Young corn seedlings contain a relative abundance.

Phytochrome is far from evenly distributed. In 6-day corn seedlings, it was found most concentrated in two areas: (1) In the higher part of the actively lengthening first internode (the stem section between joints, first springing up from the seed); and (2) in the first developing leaf, folded sheathlike around its stem.

Phytochrome in the Open

Knowing where phytochrome is and how much of it to expect at best, the research team applied fractioning processes to corn plant tissues.

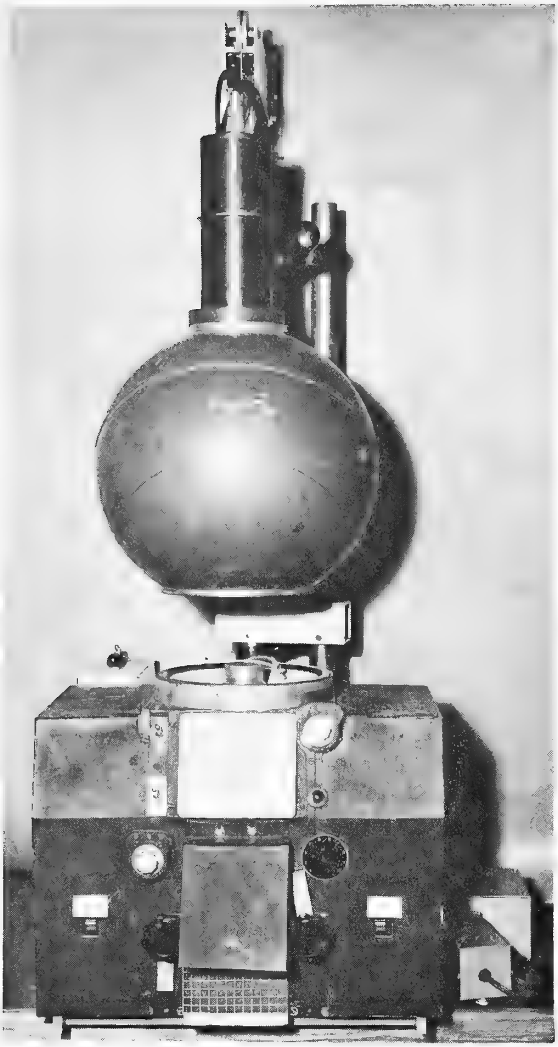

Figure 3.--An assay of phytochrome in the dual-monochromator spectrophotometer starts with centering a sample, such as a bean seedling, so that the 12-inch sphere and projecting phototube can be lowered over it. The oblong base houses the lamp for measuring pigment changes, lenses, mirrors, and other optical parts of the instrument, as well as the electrical measuring equipment. The operator applies irradiation of two pre-selected light waves alternately to the sample, and the meter readings on the dial show the change in phytochrome's form. The amount of change indicates the concentration of the pigment. Wave lengths used have generally been 6600 A. for red and 7350 A. for far red, because these are about the peaks of effectiveness for controlling the plant photoreactions.

These separation processes have yielded an extract in a test tube that is phytochrome much more concentrated than in the plant, its identity proved by its reactions in the pigment-detecting instrument. The greenish extract is only partially purified, that is, it contains some other plant proteins.

Total isolation of phytochrome is being tried for. This may be a time-consuming, arduous task. Yet isolation is an essential step if research scientists are to identify the elements and structure of the chemical molecule that is so vital a part of plant organism. And only when this information is available can they make solid progress in learning the way in which plants get or make their phytochrome and the way in which phytochrome's elements serve in enzyme reactions, such as the fat utilization that governs germination in some seeds and the coloration processes in vegetables and fruits.

Phytochrome in test tubes reacts as a soluble protein. Knowing its nature scientists can project its manner of serving plants a step forward. Apparently, the colored protein's molecular shift under red light influence enables the protein to function as an active enzyme; and this enzyme is the triggering agent that actually launches a growth change. Proteins

often serve as enzymes--transformers of cell tissue that activate life processes without themselves being used up as nutrients or cell-building materials.

For a substance so sensitive to light differences, phytochrome is remarkably durable and resistant to cold, although falling temperature retards its reversibility. Even at -70° C., phytochrome has retained a tenth of its reversible effectiveness. Cold as extreme as -180° C. has put it out of commission. However, warming restores full reversibility. Phytochrome has remained reversible after 9 months' storage at -15° C. (5° F.)

Fitting Phytochrome to Photoperiods

Ever since phytochrome has been separated from plants, one question of crucial importance has been: Does this pigment inside living plants shift its form at night at a rate in keeping with all that has been learned in 40 years about plants' dark photoperiod timing?

The light beam instruments alone did not answer this. They demonstrate a rapid reversibility of phytochrome, in living plants or as an extract, under successive irradiation with red and far-red light--but not the natural rate of change toward inactive form in darkness.

As a way of measuring this rate of change in darkness, the photoperiodic team used a light beam instrument to check the state of phytochrome in corn seedlings and other living plant tissues at different stages of each dark period. After 2, 4, and 6 hours of a dark period, living plant materials were put under far-red light to learn how much concentration of the inactive form the phytochrome would show.

This check test has tied phytochrome, as it is known today, firmly to the photoperiodic law of nature discovered by Garner and Allard. The instrument recordings show phytochrome shifting to inactive form at orderly rates during dark photoperiods--confirming that the blue pigment accounts for the clocking of darkness in fields, forests, and gardens.

A shift from active to predominantly inactive form in darkness takes on the average about 12 hours. But the ratio differs with plant species, and slows when temperature falls.

Concentration of the pigment changes faster at the onset of darkness than during the later hours of the night. After only 2 hours of darkness, a corn seedling has reached the half-point of total change. The corn plant's rate is fairly typical, judging by tests on many plants.

In nature itself, seedling tree experiments show impressively how the reversibility of phytochrome works photoperiodically, and how it can be managed. For example, a red maple springing from seed grows scarcely more than half a foot its first year, in New England where long nights prevail. In one experiment, a red maple grown from seed stood half a foot tall at 10 months when given this normal light ration. But a companion red maple grew to 8-foot height from seed in the same 10 months, and kept leaf production in pace with stem growth, because its schedule was a continuous "summer" of long days and short nights. The fast-growing tree's light allowance was adjusted so that the total energy, spread over

16-hour days, was no more than the small tree's. The one difference, a continuous round of long days and short nights, kept the fast-growing tree's phytochrome in active form at sufficient concentration to maintain non-stop growth.

Some additional kinds of young trees, though not all, evidently can be induced to grow constantly on rightly managed photoperiodic schedules. For nurserymen and plant breeders, such experiments promise methods for speeding production of slow-growing stock.

SOME ADDITIONAL CLUES AND FINDINGS

Control Centers and Transmission

In recent years, a start has been made toward locating control centers from which phytochrome operates in plants, and learning the spread effects to the growth change location. Evidence such as the following indicates that both the control centers and the transmission system may vary considerably with the kind of plant and its growth stage.

Mature leaves are known to be control centers in which phytochrome concentrates and gives flowering signals. In some cases, leaf signals are transmitted to distant flowering shoots. Perhaps the first revelation of a long-range network was the evidence that a single leaf on a soybean plant could provide adequate stimulus to launch the whole plant's flowering (page 6). Many experiments since have shown leaves doing such work.

In the pink flowering shrub Weigela, phytochrome in the uppermost pair of leaves is apparently the control center for growth in each branch. In one experiment, bud development in greenhouse Weigela was started by providing summer-length nights, then halted by lengthening nights to 12 hours--and then, the two terminal leaves on some branches were removed and these branches resumed bud growth. The artificially induced activity did not last long unless the plants were given longer days, resembling their budding season. Yet the plants had reacted as if their leaf-leaders were gone and had taken a halt signal with them. Removing lower leaves had no such effect.

Phytochrome at times operates a remarkably short-range network. Mature tomatoes require red light so directly on each part to develop the yellow in their skin that the phytochrome signals reach no more than two millimeters (about a twenty-fifth of an inch) beyond an irradiated area.

Tests have shown that the first 5 days of ripening, after tomatoes reach maturity, are the critical time in which they can launch skin coloring. Light treatment started later has no skin coloring effect. So little strength of light is needed that five-hundredths of a foot-candle--scarcely brighter than moonlight--is enough to bring out the skin color if continued an hour a day for the ripening period of 10 to 14 days.

Off-season market tomatoes do not develop the yellow pigment during dark-room ripening and storage, so that the flesh under the transparent, colorless skin shows through as pink, rather than red. The photoperiodic tests indicate that lighting and turning tomatoes in commercial ripening rooms would be necessary for uniformly bright color--a process probably too costly for wide commercial use.

Some Seeds Use Light; Others Manage Without

All seeds carry phytochrome, and many plants use it to get out of the ground. Phytochrome in some seeds needs scarcely more than a light flash. Seeds of some of the worst weeds are in this class. Farmers make use of a photoperiodic principle when they follow advice not to cultivate after applying a pre-emergence herbicide. These weed-killers do not penetrate soil deeply, but neither can deep-buried weed seed make a start without light. Turning soil up and under can give these seed light aplenty to bring up a weed crop. On the other hand, there are plant seed that require more light to assure germination.

Still other kinds of plants would not exist if their buried seed depended on light. They survive and thrive in a light-controlled world because nature has allowed an emergency hook-up in the seeds' growth-starting mechanism. Once out of the ground, these seed plants commonly follow conventional photoperiodic channels and behavior. Scientists emphasize that evasions of photoreaction in plant growth are unusual enough to be the proverbial exceptions that "prove the rule." Understanding a non-conforming plant is like understanding the normal individual from the behavior of abnormal ones.

Seed of a plant such as tobacco normally depend on light to start germination, but can manage without light provided temperature gives a go-signal. The right alternations between coolness and warmth aid these seed to sprout in darkness.

Seed of crops like corn regularly sprout without their light-signaling pigment. Although the seed receive light at planting time, the phytochrome does not react because the seed are dry. They germinate in darkness, evidently by aid of favorable moisture, temperature, and other soil conditions.

LIGHT AS PART OF THE MECHANISM

Sunlight

Since the sun pours red and far-red light at one time on plants, this raises a question: Is some difference in sunlight an original cause of earth's vegetational growth changes?

It is evident that when bright sunlight strikes plants in the open, red overinfluences the far red and pushes phytochrome's dual-form atoms toward concentration in active form. A forest cover, on the other hand, may absorb so much of sunlight's red that far red becomes the ruling influence.

In any case, when light containing both red and far red strikes phytochrome, there is some competitive pushing of the reversible atoms, which are always in motion. This is in contrast to the one-way push toward concentration during a dark photoperiod or under artificial light limited to red or far red.

Artificial Light

A basic fact about indoor light installations is that the two types of commonly used lamps differ greatly in the proportions of red and far red in their white light. Light from ordinary incandescent filament lamps contains large amounts of both red and far red, as sunlight does. Light from fluorescent lamps is high in red and extremely low in far red. In a greenhouse with fluorescent installations, if more far red is wanted, it can be obtained by adding some incandescent lamps, since plants sense and respond to such adjustments.

Both incandescent and fluorescent lamps have a useful place in artificially lighted growth rooms. For example, fluorescent installations are the better source for fulfilling high-intensity requirements. On the other hand, incandescent lamps have proved more efficient for purposes such as speeding the flowering of long-day plants and promoting swifter growth in woody and foliage plants--even though these lamps shed much of the inhibiting far red.

When separate red or far-red light is specifically wanted for indoor plant work, filters of colored cellophane can provide relatively pure color. Almost pure red can be obtained by equipping a fluorescent lamp with a red filter, because this blocks out light waves shorter than those of the red band, and the lamp itself produces a negligible amount of far red. Amost pure far red can be obtained by equipping an incandescent filament lamp with blue and red filters, because these together block out almost the entire visible spectrum, letting through the far red which such lamps can provide.

THE ROAD AHEAD

Pioneering plant scientists have gone far in exploring light-growth relationships, but still regard the interior of growing plants as a dark continent to a considerable extent. The intricacy of photoperiodic reactions is evident, even in a brief account such as this, limited to major landmark discoveries with a little experimental work cited. Actually, experiments by hundreds have contributed facts that clarify and complicate light-growth processes. World-wide scientific reports on photoperiodism are numerous.

Photoperiodic specialists have predicted that discoveries in the coming decade should outstrip past progress. From today's position, scientific research can work forward toward further goals, such as completely analyzing and describing the chemical structure of phytochrome and establishing the nature of its enzymatic action. Because phytochrome is a protein, it probably cannot be made synthetically but its enzymatic action probably can be controlled by compounds still to be discovered.

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